

TREATMENT WITH RESVERATROL DURING *IN VITRO* MATURATION IMPROVES PORCINE OOCYTE QUALITY AND EMBRYONIC DEVELOPMENT*

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Introduction

The developmental competence of somatic cell nuclear transfer (SCNT) embryos from porcine *in vitro* matured (IVM) oocytes is lower than that of *in vivo* counterparts. Insufficient cytoplasmic maturation of porcine IVM oocytes is thought to be responsible for this low efficiency. The aim of the present study was to determine whether the supplementation of resveratrol during IVM can improve porcine oocyte quality and SCNT embryo development of these oocytes.

Materials and Methods

Porcine Cumulus-Oocyte-Complexes were cultured in medium supplemented with different concentration of resveratrol (0, 0.1, 1.0, 5.0 and 10.0 μ M). At 44h post IVM, the polar body extrusion was examined to evaluate the nuclear maturation rates. H2DCFDA and 2-NBDG staining were used to assay intracellular reactive oxygen species (ROS) levels and glucose uptake capability of oocytes, respectively. Actin and tubulin staining were used to assay cytoskeleton structure. In addition, qRT-PCR was used to examine the expression of apoptosis and oxidative stress-related genes including *Bax*, *Bcl2*, *CAT* and *SOD1*. Finally, the developmental competency of parthenogenetic activation (PA) and SCNT embryos derived from IVM oocytes were used to determine whether the treatment of resveratrol during IVM could improve the subsequent embryonic development.

Results and Conclusion

The results showed that: (1) after 44h of IVM, no significant difference was observed in nuclear maturation rates of the 0.1, 1.0, and 10.0 μ M resveratrol groups (75.1%, 78.2% and 79.0% respectively) compared with that of the control group (72.6%, $P > 0.05$), but the nuclear maturation rate of 5.0 μ M resveratrol group (84.5%, $P < 0.05$) was significantly increased; (2) resveratrol treatment at the optimum concentration of 5.0 μ M significantly reduced intracellular ROS, increased the ability of glucose uptake, and improved cytoskeletal dynamics, indicating that treatment of 5.0 μ M resveratrol can improve the quality of porcine IVM oocytes; (3) there was significantly lower expression of the proapoptotic gene (*Bax*) and higher expression of the antiapoptotic gene (*Bcl2*) in oocytes with matured nucleus ($P < 0.05$), and the catalase (*CAT*), superoxide dismutase 1 (*SOD1*) mRNA gene expression were significantly higher than that in the control group ($P < 0.05$); (4) blastocyst formation rate (15.2% vs 12.5%, $P < 0.05$) and total cell numbers (37.7 vs 30.5, $P < 0.05$) of 5 μ M resveratrol treated IVM oocytes group after SCNT were significantly higher than the control group. Collectively, our results showed that treatment with 5.0 μ M resveratrol during IVM could improve porcine oocyte quality by decreasing intracellular ROS level, increasing glucose uptake capability and regulating gene expression, and then enhancing subsequent embryo development.

*This study was jointly funded by Guangxi special project for culturing academic and technical leaders of New Century (Year 2016) and the Open Project of Guangxi Key Laboratory of Livestock Genetic Improvement (2017GXKLLGI-01).